

印度蛇菰的三萜成分*

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摘要 从我国民间药用植物印度蛇菰 (*Balanophora indica* (Am.) Griff.) 中分离得到 7 个化合物, 经鉴定为: 棕榈酰 β -香树酯(蛇菰素 A)、棕榈酰羽扇豆烯醇酯(蛇菰素 B)、乙酰 β -香树酯、乙酰羽扇豆烯醇酯、 β -香树脂酮、羽扇豆烯酮及棕榈酸。运用光谱和化学的方法对它们的结构进行解析。其中羽扇豆型萜为首次自该植物中分得; 蛇菰素 A 和 B 具有较强的护肝作用。

关键词 印度蛇菰, 三萜酯, 蛇菰素 A, 蛇菰素 B

分类号 Q 946

Triterpene Constituents from *Balanophora indica*

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Abstract From herbs of the Chinese folk medical plant *Balanophora indica*, seven compounds: β -amyirin palmitate (balanophorin A), lupeol palmitate (balanophorin B), β -amyirin acetate, lupeol acetate, β -amyrone, lupeone and palmitic acid were isolated. Their structures were elucidated by the basis of spectral and chemical evidences, respectively. The compounds of lupane were obtained from *B. indica*, firstly. Balanophorin A and B exhibited strong activity against liver damage induced by CCl_4 .

Key words *Balanophora indica*, Triterpene ester, Balanophorin A and B

Balanophora indica (Am.) Griff. was a kind of parasitic plant with root, belong to species of the family Balanophoraceae. It is used as a folk medicine plant for the tonics and hemostatic, indigenous to the province Yunnan. To our knowledge, the biologically active principle and constituents from this plant, have not been described as yet. Therefore we studied on the constituents of it. Here we report on the investigation of the isolation and structural elucidation of two triterpene esters from this plant.

RESULTS AND DISCUSSION

The petroleum benzine extract of the herbs of *Balanophora indica* (Am.) Griff, was subjected to repeated column chromatography on silica gel and aluminum oxide, to give 7 compounds: β -amyirin 3-palmitate (1, balanophorin A), β -amyrone (2), β -amyirin 3-acetate (3), lupeol 3-palmitate (4, balanophorin B), lupeone (5), lupeol 3-

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acetate (6) and palmitic acid (7). The yield of balanophorin A and B were 1.21% and 1.34%, respectively.

Balanophorin A showed a molecular ion peak at m/z 665 ($M+1$, $C_{46}H_{80}O_2$) in the mass spectrum. The IR spectrum showed at 1710, 1165, and haven't at 3300–3500 cm^{-1} , indicating the presence of a carbonyloxy group ($O=C-O$). The 1H -NMR spectrum appeared one olefinic proton signal at δ 5.15 (1H, brs), one oxygen-bearing carbon proton signal at δ 4.47 (1H, br, t $J=7.4$ Hz) in the lowfield range; and nine methyl proton at δ 1.11 (3H, s), 0.94 (3H, s), 0.93 (3H, s), 0.84 (15H, brs, $5 \times CH_3$), 0.80 (3H, s); seventeen methylene protons at δ 1.23 (34H, brs, $17 \times CH_2$), and the great majority protons signal showed gather at δ 0.80–2.28 rang in the highfield. It suggested that balanophorin A could be an triterpene ester derivative. The IR spectrum absorption at 1370, 1350, were indicated that balanophorin A could be belong to the β -amyrin type triterpene ester derivative (Snatzke *et al*, 1962). The ^{13}C NMR and DEPT spectrum of balanophorin A revealed one carbonyl carbon (δ 173.4), nine methyl carbons (δ 24.3, 16.8, 25.9, 28.0, 32.4, 23.7, 14.1), six quaternary carbons (δ 38.2, 39.8, 37.1, 41.7, 32.6, 34.8), four methine carbon [δ 80.5 (bearing an oxygen atom), 55.2, 47.5, 47.2] and two olefinic carbons (δ 121.6d, 145.1s). These data can be accommodated on the β -amyrin type triterpene having long chain fatty acid. The EI-mass spectrum of balanophorin A showed a characteristic fragment ion for the loss of palmitic acid at m/z 218 (base peak). On comparison of the ^{13}C NMR and 1H -NMR spectrum of balanophorin A with that of β -amyrin (Bhattacharyya *et al*, 1986), was identified as β -amyrin 3-palmitate (1). The EI-mass spectrum of the alkaline hydrolysis product was corresponding with that of β -amyrin, and further confirmed that balanophorin A should be assigned to the β -amyrin ester. From the above evidence, the structure of balanophorin A was established to be β -amyrin 3-palmitate.

Balanophorin B was calculated for $C_{46}H_{80}O_2$ by the FABMS and ^{13}C NMR spectrometry. The IR spectrum appeared two absorption at 1710 ($C=O$), and 1625 ($C=C$). The 1H NMR spectrum was quite similar to that of balanophorin A, but it showed two olefinic proton signals at 4.66 (1H, s), 4.54 (1H, s); eight methyl proton at δ 0.76 (3, s), 0.81 (9H, s, $3 \times CH_3$), 0.84 (3H, s), 0.92 (3H, s), 1.00 (3H, s), 1.66 (3H, s); and seventeen methylene protons at δ 1.23 (34H, brs, $17 \times CH_2$). The ^{13}C NMR and DEPT spectrum revealed one carbon (δ 173.5), two olefinic carbons (δ 109.4t, 150.8s), five quaternary carbons (δ 38.9, 4.09, 38.1, 42.7, 43.0), and six methine carbon (δ 80.7, 55.5, 50.4, 37.2, 48.4, 48.0). It expressed that balanophorin B could be a lupeol type derivative. The EI-mass spectrum of it exhibited a characteristic fragment ion for the loss of palmitic acid at m/z 409 ($M-256$), 239, 257, and 426. It was identified as lupeol 3-palmitate (4) by comparison of the data of NMR with lupeol (Dreyer *et al*, 1972). Alkaline hydrolysis of it yields a triterpene compound. The EIMS of the triterpene was corresponding with that of lupeol. The structure of it was established to be lupeol 3-palmitate, again. This compound was obtained from *B. indica*, firstly (Yadagiri *et al*, 1984; Chengalur *et al*, 1976).

Balanophorin A and B exhibited strong activities against liver damage induced by CCl_4 in mice (Lin *et al*, 1988).

EXPERIMENTAL

General procedures The NMR spectra were performed chloroformed using TMS as int. standard at 400MHz with a Bruker AM-400 instrument. The carbon type was determined by DEPT experiments. IR spectra were recorded in KBr pellets on a Perkin-Elmer 577 interferometer. EIMS and FABMS: positive, direct inlet 70eV on VG Autospec instrument. For CC, silica gel (200–300 mesh, Qingdao) and aluminum oxide (neutral, 200–300 mesh, Shanghai). TLC precoated silic gel plate HF_{254} (0.25mm in thickness).

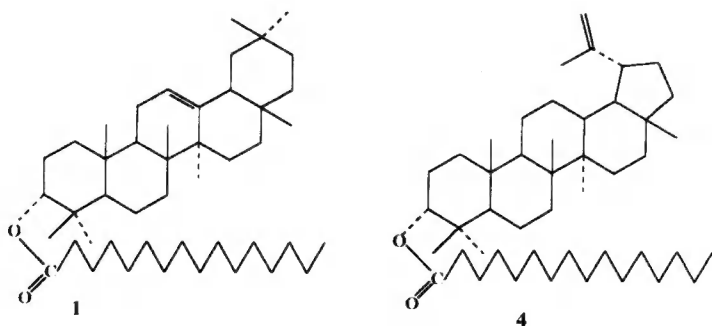
Plant material The whole plant of *Balanophora indica* (Am.) Griff were collected in Xishuangbanna, Yunnan province, in October, 1996, and identified by Mr. Cui Jing-yun, Xishuangbanna Tropical Botanic Garden, the Chinese Academy of Sciences, where the herbarium specimen has been deposited.

Identification of the known triterpenes All of the known were identified by comparison with authentic samples by their NMR or MS spectra data.

Extraction and Separation of triterpenes The dry whole plant powered material (700 g) was extracted with petroleum benzene yielding, after evapn, a brown yellow oil residue (80 g). The petroleum benzene extract was dissolved in benzene and extracted with methanol. The benzene layer, on evapn of the solvent, 46 g residue was obtained. The benzene extract chromatographed on a column of silica gel with petroleum benzene - acetone (20:1 ~ 2:1) to give 3 fractions in increasing of polarity. Fraction 2 (29 g) was purified by CC on aluminum oxide (neutral) with benzene - methanol (4:1) to furnish balanophorin A (8.5 g) and B (9.4 g). Fraction 3 was separated similarly as that for fraction 2 to afford β -amyryn 3 - acetate (40 mg), β -amyrone (21 mg). And lupeol 3 - acetate (34 mg), lupeone (25 mg). Palmitic acid was obtained from fractoin 1.

Balanophorin A (1). Amorphous white powders (Me_2CO), mp 77°C , dissolved in chloroform, petroleum benzene and benaene. IR $\nu_{\text{max}}^{\text{KBr}} \text{cm}^{-1}$: 2900, 2825, 1710, 1450, 1370, 1350, 1235, 1165, 1085, 980, 710. FABMS (m/z): 665 ($M+1$). EIMS (m/z): 665, 409, 218 (base peak), 257, 239, 203, 190, 175, 109, 69. ^1H NMR (CDCl_3): δ 5.15 (1H, brs, 12-H), 4.47 (1H, t, $J=7.4\text{Hz}$, 3-H), 2.26 (2H, t, $J=7.4\text{Hz}$, 2'-H), 1.23 (34H, brm, CH_2), 1.11 (3H, s, 27- CH_3), 0.94 (3H, s, 30- CH_3), 0.93 (3H, s, 29- CH_3), 0.84 (15H, brs, CH_3), 0.80 (3H, s, 16'- CH_3). ^{13}C NMR data see table. Balanophorin A (20 mg), was treated with NaOH - H_2O (2.0 g NaOH dissolved in 20 ml H_2O) at 160°C for 12 hr. The reaction mix was extracted with CHCl_3 , after evapn, obtained a residue. The EIMS spectra of the residue exhibited a fragment ion peak at m/z 426, 218 (base peak).

Balanophorin B (4). Amorphous white powders (Me_2CO), mp $68 \sim 69^\circ\text{C}$. IR $\nu_{\text{max}}^{\text{KBr}} \text{cm}^{-1}$: 3040, 2900, 2825, 1710, 1625, 1450, 1440, 1438, 1370, 1165, 970, 900, 710. EIMS (m/z): 665 ($M+1$), 409, 426, 257, 239, 218, 95, 68, 55 (base peak). ^1H NMR (CDCl_3): δ 4.46 (1H, s, 29-H), 4.54 (1H, s, 29-H), 4.44 (1H, dd, $J=10.5, 5.6\text{Hz}$, 3-H), 1.23 (34H, brm, CH_2), 1.66 (3H, s, 30- CH_3), 1.00 (3H, s, 27- CH_3), 0.92 (3H, 24- CH_3), 0.84 (3H, s, 29- CH_3), 0.81 (9H, s), 0.76 (3H, s), 0.76 (3H, s, 16'- CH_3), 2.26 (2H, t, $J=7.6\text{Hz}$, 2'- CH_2), 2.31 (1H, m, 18-H). ^{13}C NMR data see table. Balanophorin B (20 mg), was treated with NaOH - H_2O (2.0 g NaOH dissolved in 20 ml H_2O) at 160°C for 12 hr. The EIMS spectra of the product exhibited a fragment ion peak at m/z 426, 189, 203.



β -Amyrone (2). $\text{C}_{30}\text{H}_{48}\text{O}$, M424. Colorless needles (MeOH). The EIMS, ^1H and ^{13}C - NMR spectra data were identical to the published reference spectra data of β -amyrone (Gonzalez *et al*, 1981).

β -Amyryn acetate (3). $\text{C}_{32}\text{H}_{52}\text{O}_2$, M468. Colorless pillars (Me_2CO), mp $236 \sim 237^\circ\text{C}$. Similarly, the EIMS ^1H and ^{13}C - NMR spectra data were identical to those of the reference compound β -amyryn acetate (Seo *et al*, 1975).

Lupeone (5). $\text{C}_{30}\text{H}_{48}\text{O}$, M424. White needles (MeOH). The EI - MS, ^1H and ^{13}C - NMR spectra were corresponding with that of lupeone 3 - one (Budzikiewicz *et al*, 1963).

表 1 蛇菰素 A 和 B 的 ^{13}C NMR 数据
Table 1 ^{13}C NMR data of balanophorin A and B

C (碳序)	Balanophorin A 蛇菰素 A	Balanophorin B 蛇菰素 B	C (碳序)	Balanophorin A 蛇菰素 A	Balanophorin B 蛇菰素 B
1	37.7 t	37.8 t	21	31.9 t	29.4 t
2	23.6 t	23.4 t	22	36.8 t	40.0 t
3	80.5 t	80.7 t	23	28.4 q	27.5 q
4	38.2 s	38.54 s	24	16.8 q	16.3 q
5	55.2 d	55.5 d	25	15.5 q	16.3 q
6	18.2 t	18.3 t	26	16.8 q	16.3 q
7	33.3 t	34.3 t	27	25.8 q	14.5 q
8	39.8 s	40.9 s	28	28.0 q	18.3 q
9	47.5 d	50.4 d	29	32.4 q	109.4 t
10	37.1 s	38.1 s	30	23.7 q	19.4 q
11	23.5 t	21.0 t	1'	173.4 s	173.5 s
12	121.6 d	25.2 t	2'	34.9 t	34.8 t
13	145.1 s	37.2 d	3'	25.1 t	25.2 t
14	41.7 s	42.7 s	4'	29.3 t	29.3 t
15	26.9 t	27.5 t	⋮	⋮	⋮
16	26.2 t	35.6 t	⋮	⋮	⋮
17	32.6 s	43.0 s	13'	29.7 t	30.8 t
18	47.2 d	48.4 d	14'	31.0 t	31.9 t
19	46.8 t	48.0 d	15'	22.7 t	22.7 t
20	34.8 s	150.8 s	16'	14.1 q	14.5 q

Measured in CDCl_3 , ppm.

Lupeol acetate (6), $\text{C}_{32}\text{H}_{52}\text{O}_2$, M468. Colorless pillars (MeOH), mp 217 ~ 218°C. The EIMS and NMR spectra data were identical to those of the reference compound lupeol 3 - acetate (Wenkert *et al*, 1978).

Palmitic acid (7), $\text{C}_{16}\text{H}_{32}\text{O}_2$, M256. Colorless powders (MeOH). The EIMS and NMR spectra were corresponding with that of palmitic acid (Vanderlan *et al*, 1991).

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名次	期刊名称	被引频次	名次	期刊名称	影响因子
1	植物学报	943	1	植物生态学报	0.4556
2	植物生理学通讯	553	2	植物学报	0.4286
3	植物生理学报	489	3	遗传学报	0.3926
4	遗传学报	419	4	植物生理学报	0.3860
5	生物化学与生物物理进展	383	5	古脊椎动物学报	0.3265
6	植物分类学报	366	6	细胞生物学杂志	0.2857
7	生物化学与生物物理学报	323	7	微生物学报	0.2549
8	云南植物研究	308	8	植物分类学报	0.2500
9	古植物学报	266	9	人类学学报	0.2444
10	动物学报	264	10	实验生物学报	0.2397

1. 数据来源:中国科学院文献情报中心中国科学引文数据库 1996 年数据。本表由中国科学引文数据库统计编制。
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